EVALUATION OF NEW MANAGEMENT APPROACHES AGAINST LEMON BUTTERFLY (*PAPILIO DEMOLEUS* L.) INFESTING JARA LEMON IN SYLHET, BANGLADESH

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ABSTRACT

The study was conducted at Bagerkhal village under Fatehpur union, Jaintapur, Sylhet, Bangladesh during April to September, 2016 to determine the most effective new management approaches in suppressing lemon butterfly larvae infesting leaves of Jara lemon plant. Five approaches viz., handpicking and killing of lemon butterfly larvae (T₁), spraying of imidacloprid (Sapta 70 WG) @ 0.5 g L⁻¹ of water (T₂), bioneem plus (Azadirachtin 1 EC) @ 1 ml L⁻¹ of water (T₂), spinosad (Tracer 45 SC) @ 1.25 ml L⁻¹ of water (T₄), chlorfenapyr (Proclaim 5 SG) @ 1 g L⁻¹ of water (T_s) and an untreated control (water spray only) (T_c) were evaluated. The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications. Approach with insecticides was applied four times at 20-day intervals. Eight plants were selected and tagged at random to collect leaf infestation data from east, west, north and south canopies of the tagged plants. Leaf infestation (%) of the whole plant was calculated by the mean no. of infestation recorded from east, west, north and south canopies of the plant. The lowest leaf infestation (17.83%) was recorded in plants treated with spinosad (T_A) and chlorfenapyr (T_c) . The highest leaf infestation (31.83%) was recorded in untreated control plants. The intermediate leaf infestation (20.85% to 21.33%) was recorded in plants treated with hand picking and killing of lemon butterfly larvae, imidacloprid and bioneem plus. Spinosad and chlorefenapyr treated plants produced the highest fruit yield (1.35 t ha⁻¹) and also gave the highest financial returns in terms of Marginal Benefit Cost Ratio (MBCR) (11.50: 1.0). The lowest MBCR (7.31: 1.0) was obtained from plants treated with hand picking and killing of lemon butterfly larvae. The results clearly indicated that spinosad is the most cost-effective as well as environmentally safe management approach for lemon butterfly larvae.

Keywords: Jara Lemon, lemon butterfly, handpicking, approach with biopesticide, MBCR.

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INTRODUCTION

Citrus trees are grown in all over Bangladesh, but majority of which are grown well in Sylhet region. Rutaceae family of lemon is one of the most common citruses. Probably, the lemon is native to North-Eastern India, Northern Burma and China. Jara lemon tree (Citrus pennivesiculata L.), commonly known as citrone is short, 3-5 m high with stout stiff thorns, petioles are short margined having a distinct articulation but not winged. Annual citrus production of the world increased @ 2.3% during 1992 to 2002 with about 13.7 million tons of lemon and limes (Yara 2017). The fruits are known as sour fruits of the world which have high nutritional importance. The most famous citrus fruits are lemon, limes, oranges, mandarin, etc. Citrus fruits being main sources of vitamin C have distinct aroma and delicious taste (Biolatto et al. 2005, Sfgate 2017). Citrus fruits are the sources of carbohydrates like sucrose, glucose, fructose and dietary fiber. They decrease circulating blood cholesterol and having a fruit after a meal improves iron absorption (BIRDEM 2013). In Bangladesh, average eating of vitamin C is far below than standard health requirements and thus 93% of the total people are suffering from various health problems due to lack of vitamin C intake. Therefore, eating fresh lemon can greatly contribute to the improvement of human health (BIRDEM 2013). The citrus fruits contain some organic compounds which work against asthma, antidepressant, stress relief, aids digestion, colds, flu, fever, nose-bleeding, mouth ulcers, throat infection and boils (Sfgate 2017). A recent study has shown that citron contains some cardiovascular benefits too (Klein 2014). Fruits are sour in taste and commonly used in preparing juice, squash, citric acid, pectin, peel, oil jam, jelly, pickle and salad. Fruit peels are used in industry to make furniture polishing material, detergents, soaps and shampoos, perfume blending, etc.

Commercial and homestead cultivation of lemon fruits could considerably improve livelihood of the rural people in Bangladesh (Sarker *et al.* 2017). Despite the favorable climatic conditions for year-round citrus production in the country, its production has been facing many problems such as insect pest infestation, postharvest losses, glut in peak season and information gap in domestic and export markets (Hortex Foundation 2010). Among these problems, insect pest has been regarded as a major problem of Jara lemon production in Bangladesh (Bhuiyan *et al.* 2016, Ehsanullah *et al.* 2017). In India, citrus plants are attacked by more than 250 insect species at various stages of plant growth from nursery to main orchards. About 165 species cause severe damage to citrus plants incurring around 30% yield loss (Vattikonda and Sangam 2017). Among the insect pests of citrus, the genus *Papilio* is widely distributed all over the world and other species of citrus butterfly occur in different parts of the world. The species *Papilio demoleus* L. being known

as lemon butterfly is the most prevalent species and mostly found in greater parts of Asia, Japan and Formosa Island in Taiwan(Sarada *et al.* 2014). Very recent study stated that lemon butterfly larvae are the most severe pest of young citrus plants in Sylhet region (Bhuiyan *et al.* 2016, Ehsanullah *et al.* 2017). The attack of lemon butterfly on citrus leaves directly affects the growth and development of citrus plant and fruits. Lemon butterfly is one of the economically important pests whose larval forms cause serious damage to the wild and cultivated species of citrus during the later stages of development. The larvae may cause complete defoliation of infested young plants (Butani and Jotwani 1975). It is a major pest in nurseries, young seedlings and new flush in mature citrus trees. It causes 83% defoliation in young citrus plants (Narayanamma *et al.* 2001). The caterpillars feed voraciously and cause extensive damage to nurseries and young seedlings leaving only midribs.

In recent years, application of conventional chemical insecticides has not been proved to be an appropriate management strategy due to high cost, development of pesticide resistance, killing of natural enemies and non-target organisms, residue in food, killing of wild lives, aquatic lives and pollution of ground water, air and other elements of nature (FAO 2016). Considering these harmful effects by synthetic chemical pesticides, some new ecofriendly management approaches including insecticides (i.e., easily bio-degradable, environmentally safe, water soluble, less persistent, less risk to apply in presence of natural enemies, more toxic to pests compared to conventional pesticides) need to be evaluated for suppressing lemon butterfly larvae. Although several researchers have evaluated spinosad, imidacloprid and chlorfenapyr against major insect pests (whitefly, thrips, brinjal shoot and fruit borer, tomato fruit borer, mango hopper, litchi mite etc.) of fruits and vegetables (Kadam et al. 2014, Jadhav et al. 2017, Sahu et al. 2018, Ranjan et al. 2019), little works have been done to develop ecofriendly management approaches against lemon butterfly larvae using these new safer insecticides. Considering the above circumstances, some new ecofriendly management approaches with insecticides were evaluated in this study to develop environmentally safe and effective management techniques against lemon butterfly larvae infesting Jara lemon in Sylhet region.

MATERIALS AND METHODS

Study site and citrus garden: The experiment was conducted at Bagerkhal village under Fatehpur union, Jaintapur under the Sylhet district of Bangladesh during April to September, 2016. Because leaf infestation of lemon butterfly larvae mostly occurs in nurseries and young citrus plants, 4-year old Jara lemon garden was selected to evaluate the efficacy of different new ecofriendly IPM approaches including safer insecticides and hand picking &killing of lemon butterfly larvae. The experimental

site is located at East-Northern corner of Bangladesh lying between 23057' and 25013' North latitude, 90056' and 92021' East longitude. The site falls under the Agroecological Zone-20: Eastern Surma-Kusiyara Flood Plain (UNDP and FAO 1988). The study area is medium high land, fertile and well drained. The citrus cultivation is mostly confined to high land, medium high land and medium land (BBS 2013). The soil of Jaintapur is acidic in reaction with pH of 5.4 (SRDI 2015).

Design of experiment and treatment application: The experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications. Six treatments viz., hand picking and killing of lemon butterfly larvae (T_1) , spraying of imidacloprid (Sapta 70 WG) @ 0.5 g L⁻¹ of water (T_2) , bioneem plus (Azadirachtin 1 EC) @ 1 ml L⁻¹ of water (T_3) , spinosad (Tracer 45 SC) @ 1.25 ml L⁻¹ of water (T_4) , chlorfenapyr (Intrepid 10 SG) @ 1 g L⁻¹ of water (T_5) and untreated control (water spray only) (T_6) were applied on Jara lemon trees. The treatments were randomly allotted to each replication. The insecticides were sprayed on the trees with a Knapsack sprayer. Trees of each plot were examined thoroughly at 20-day intervals to record the number of infested leaves.

Data collection: Random sampling techniques were used to collect data. Insect pest status on the trees of study sites were determined by determining their infestation on tree leaves. For primary data, a total of 18 trees were selected randomly. Then 4 canopies were selected from one tree of 4 canopy directions (north, east, west and south canopies) and tagged with four different colors. The numbers of infested leaves were counted at 20-day intervals. The infestation rate was calculated from the mean data of three replicates for each treatment. A total of 18 trees (3 trees per treatment) were selected at random for data collection. Then four canopies from north, east, west and south sides of the selected tree were tagged with four different colors. Yellow tape was used to tag the selected trees and binding paper was used to put number on the selected trees. The number of infested leaves were counted and transformed into percentage from means of the three replicates for each treatment. The numbers of infested leaves were counted from 50 leaves and then calculated leaf infestation in percentage. At the end of the season, the quantity of harvested marketable healthy Jara lemon fruits (t ha⁻¹) produced from the tree of each treatment including the untreated control was quantified. The costs incurred due to labor, insecticides, application of insecticides and all other materials were recorded during the study. The untreated control did not require any pest management cost. The costs for each treatment and that of untreated control were calculated based on the local market price.

Economic analysis: Marginal Benefit Cost Ratio (MBCR) was calculated considering the total expenditure of management cost and the total return from that particular treatment as recommended by Hossain *et al.* (2016) as follows:

$$MBCR = \frac{Adjusted net return}{Total management cost}$$

In this study, the cost was calculated by adding all costs incurred due to labors, insecticides and application of insecticides for each treatment including control during the entire study period from April to August 2016. The yield from each treatment in terms of taka was calculated by multiplying the total yield and the unit price of jara Lemon (80 Tk kg⁻¹). Net return was calculated by subtracting the treatment wise management cost from gross return. The adjusted net return was determined as shown below:

Adjusted net return = Net return from the treated plant - Net return from the control plant.

Statistical analysis: The collected data on infested leaves during study were tabulated and statistically analyzed using R package software (R Development Core Team 2016). The data of all quantitative characters were statistically analyzed to find out the statistical significance of the experimental results. The means of all the treatments were calculated and analysis of variance (ANOVA) for all the characters under consideration was performed by F-variance test (Gomez and Gomez 1984). Means of the treatments were compared by Least Significance Difference (LSD) test.

RESULTS

Leaf infestation increase before first treatment application: In east canopy, leaf infestation before 60 days of the first spray was 9.33% which increased up to 19.44% before 20 days of the first spray, showing 10.11% increase in leaf infestation within 40 days (Table 1). In west canopy, leaf infestation before 60 days of the first spray was 10.89% which increased up to 18.78% before 20 days of the first spray, showing 7.89% increase in leaf infestation within 40 days. In north canopy, leaf infestation before 60 days of the first spray was 11.22% which increased up to 19.73% before 20 days of the first spray, showing 8.51% increase in leaf infestation within 40 days. In south canopy, leaf infestation before 60 days of the first spray was 12.89% which increased up to 17.33% before 20 days of the first spray, showing 4.44% increase in leaf infestation within 40 days. Overall, leaf infestation before 60 days of the first spray was 11.08% in whole tree which increased up to 18.82% before 20 days of the first spray, showing 7.74% increase in leaf infestation within 40 days.

Table 1. Leaf infestation increase (%) in Jara lemon before first treatment application

Canopies/		Leaf infestation (%))	Increase in leaf
whole plant	Before 60 days of 1st spray	Before 40 days of 1stspray	Before 20 days of 1stspray	infestation within 40 days (%)
East canopy	9.33	14.78	19.44	10.11
West canopy	10.89	15.33	18.78	7.89
North canopy	11.22	16.22	19.73	8.51
South canopy	12.89	15.33	17.33	4.44
Overall leaf infestation	11.08	15.42	18.82	7.74

Percentage of leaf infestation after four different sprays: At twenty days after the 1stspray, leaf infestation did not significantly vary in all canopies except south canopy (Table 2). At twenty days after the 2nd, 3rd and 4thspray, leaf infestation varied significantly in all canopies. At twenty days after the 2nd spray, all the treatments viz., hand picking and killing of lemon butterfly larvae (T₁), spraying of imidacloprid (Sapta 70 WG) @ 0.5 g L⁻¹ of water (T₂), bioneem plus (Azadirachtin 1 EC) @ 1 ml L⁻¹ of water (T₂), spinosad (Tracer 45 SC) @ 1.25 ml L⁻¹ of water (T₄) and chlorfenapyr (Intrepid 10 SG) @ 1 g L⁻¹ of water (T_c) significantly reduced the leaf infestation over the untreated control. At twenty days after the 3rd and 4thspray, the two treatments viz., (Tracer 45 SC) @ 1.25 ml L⁻¹ of water (T₄) and chlorfenapyr (Intrepid 10 SG) @ 1 g L⁻¹ of water (T_s) most effectively reduced the leaf infestation. At twenty days after the 3^{rd} and 4^{th} spray of spinosad (T_4) and chlorfenapyr (T_5) , leaf infestation was found as the lowest level ranging from 16.7% to 17.3% in east canopy, 17.3% to 18.0% in west canopy, 18.0% to 19.3% in north canopy and 18.0% to 20.7% in south canopy. At twenty days after the 3rd and 4th spray of imidacloprid (T_2) and bioneem plus (T_2) and hand picking and killing of larvae (T_1) , leaf infestation was found as the intermediate level ranging from 20.7% to 22.7% in east canopy, 21.3% to 22.7% in west canopy, 19.3% to 22.0% in north canopy and 20.0% to 22.7% in south canopy. At the time of 3rd and 4th spray, leaf infestation was found as the highest level ranging from 26.7% to 30.7% in east canopy, 29.3% to 32.0% in west canopy, 28.7% to 31.3% in north canopy and 29.3% to 33.3% in south canopy. Similar trend in reduction of leaf infestation was also found in whole treated tree. The lowest leaf infestation (17.83%) was recorded in plants treated with spinosad (T_a) and chlorfenapyr (T_a). The highest leaf infestation (31.83%) was recorded in untreated control trees. The intermediate level of leaf infestation was found from hand picking and killing of larvae (20.85%), spraying of imidacloprid (21.50%) and bioneem plus (21.33%).

Percentage of leaf infestation in Jara lemon crop in east, west, north and south canopies of tree at 20 days after the first, second, third and fourth spraying of treatments in Sylhet during April to September, Table 2.

Leaf infestationat 20 days after each sprayin east canopy (%)	Leaf infestation at 20 days after each sprayin west canopy (%)	20 days after canopy (%)	r each	Leafin	Leaf infestation at 20 days after each sprayin north canopy (%)	0 days after canopy (%)	each	Leaf in	festation at orayin south	Leaf infestation at 20 days after each sprayin south canopy (%)	r each	Whole treated
4th 1st	2 nd	3rd	4 th	Ist	2 nd	3rd	4 th	İst	2 nd	3rd	4th	plant
20.7 18.0 2ı (4.55)b (4.24) (4.24)	20.0 (4.47)b (22.7 (4.76)b	22.0 (4.69)b	16.0 (4.00)	17.3 (4.16)c	19.3 (4.39)c	20.7 (4.55)b	15.3 (3.99)b	20.7 (4.54)b	22.0 (4.69)b	20.0 (4.47)b	20.85 (4.57)b
22.7 17.3 (4.76)b (4.16) (4.76)	19.3 (4.39)b (•	22.0 (4.69)b	21.3 (4.62)b	17.3 (4.16)	20.0 (4.47)b	22.0 (4.69)b	20.7 (4.55)b	16.0 (3.99)b	20.7 (4.54)b	22.7 (4.76)b	21.3 (4.62)b	21.50 (4.64)b
22.0 18.0 (4.76)b (4.24) (6	19.3 (4.39)b	22.7 (4.76)b	21.3 (4.62)b	18.0 (4.24)	21.3 (4.54)b	22.0 (4.69)b	20.7 (4.55)b	16.7 (4.08)b	17.3 (4.16)c	22.7 (4.76)b	21.3 (4.62)b	21.33 (4.62)b
16.7 18.7 (4.08)c (4.32) (4	19.3 (4.39)b	18.0 (4.24)c	17.3 (4.16)c	16.7 (4.08)	18.0 (4.24)c	18.7 (4.32)c	19.3 (4.39) bc	18.0 (4.24) ab	20.0 (4.47)b	20.7 (4.54)c	18.0 (4.24)c	17.83 (4.22)o
17.3 18.0 (4.16)c (4.24) (18.7 (4.31)b (18.0 (4.24)c	18.0 (4.24)c	18.0 (4.24)	20.0 (4.47)b	18.7 (4.32)c	18.0 (4.24)c	16.7 (4.08)b	17.3 (4.16)c	20.7 (4.54)c	18.0 (4.24)c	17.83 (4.22)c
30.7 19.3 2 (5.54)a (4.39) (4	24.0 (4.90)a	29.3 (5.42)a	32.0 (5.66)a	18.7 (4.31)	24.0 (4.90)a	28.7 (5.35)a	31.3 (5.60)a	20.0 (4.47)a	24.7 (4.97)a	29.3 (5.42)a	33.3 (5.77)a	31.83 (5.64)a
0.24 0.25	0.21	0.19	0.17	0.35	0.13	0.17	0.25	0.33	0.23	0.14	0.15	1.38
sn **		*	*	NS	*	*	*	*	*	*	* *	*
2.82 3.27	*	t	1 00	12.4				ç	6	-		4 10

Note: Data in the parentheses are square root transformed values. T₁ = Hand picking and killing of larvae, T₂ = Spraying of Imidacloprid (Sapta 70 WG) @ 0.5 g/L of water, T_3 = Spraying of Bioneem plus (Azadirachtin 1 EC) @ 1 ml/L of water, T_4 = Spraying of Spinosad (Tracer 45 SC) @ 1.25 ml/L of water, T_5 = Spraying of Chlorfenapyr (Intrepid 10 SG) @ 1 g/L of water and T_6 = Untreated control (spraying of water only).

Cost Benefit Analysis: Marginal Benefit Cost Ratios (MBCRs) of various treatments are presented in Table 3. In this study, the untreated control (T₆) did not require any pest management cost. Even though the variation in healthy fruit yield differed significantly among various treatments, the calculated MBCRs provided separate scenariosdue to some variations in pest management costs. The adjusted net return was the highest in trees sprayed with spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid 10 SG). The calculated MBCR for each treatment revealed that the highest MBCR (11.50) was obtained from the trees which were sprayed with spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid 10SG) and the lowest MBCR (7.31) was obtained from trees treated with hand picking and killing of lemon butterfly larvae. The calculated MBCR was found 10.43in imidacloprid (Sapta 70 WG) sprayed trees and 9.80in bioneem plus (Azadirachtin 1 EC) sprayed trees.

Table 3. Economic analysis of some new management options including biopesticides

Treatment	No. of insecticide spray	Pest management cost (Tk.)	Yield (t ha ⁻¹)	Gross return (Tk.)	Net return (Tk.)	Adjusted net return (Tk.)	MBCR (Tk./Tk.)
T_1	-	950	1.20	96000	94950	6950	7.31
T_2	4	1400	1.30	104000	102600	14600	10.43
T_3	4	1500	1.30	104000	102700	14700	9.80
T_4	4	1600	1.35	108000	106400	18400	11.50
T_5	4	1600	1.35	108000	106400	18400	11.50
T_6	-	-	1.10	88000	-	-	

Note: T_1 = Hand picking and killing of lemon butterfly larvae, T_2 = spraying of imidacloprid (Sapta 70 WG) @ 0.5 g L⁻¹ of water), T_3 = Spraying of bioneem plus (Azadirachtin 1 EC) @ 1 ml L⁻¹ of water, T_4 = Spraying of spinosad (Tracer 45 SC) @ 1.25 ml L⁻¹ of water, T_5 = Spraying of chlorfenapyr (Proclaim 5 SG) @ 1 g L⁻¹ of water, and T_6 = untreated control (water spray only). MBCR = Marginal Benefit Cost Ratio

Cost of insecticides: Sapta 5 SG, Tk 25 per 2g packet @ 0.5 g L⁻¹ of water; 2 g insecticide needed for a single spray, Azadirachtin 1EC, Tk 500 per 20 ml bottle @ 1 ml L⁻¹ of water; 5 ml insecticide needed for a single spray, Tracer 45 SC, Tk 205 per 10 ml bottle @ 1.25 ml L⁻¹ of water; 2.5 ml insecticide needed for a single spray, Proclaim 5 SG, Tk 100 per 10g packet @ 1g L⁻¹ of water; 5 g insecticide needed for a single spray. Labor cost: @ Tk 350 day⁻¹; Market price of 1 kg Jara lemon = Tk 80

DISCUSSION

Three initial leaf infestation data were recorded from 60 to 20 days before the first spray application. The rate of leaf infestation increased gradually during this period. The five different new management approaches including biopesticidesalong with handpicking and killing of lemon butterfly larvae were applied to suppress the lemon butterfly larvae. It was evident from the results that spinosad (Tracer 45SC) (T₄) and chlorfenapyr (Intrepid10 SG) (T₅) proved to be the most effective in suppressing leaf infestation in Jara lemon. Among the whole treated trees, the lowest percentage of leaf infestation (17.83%) was observed in the trees sprayed with spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid 10 SG). In addition to leaf infestation reduction, spinosad and chlorfenapyr treated trees also gave the highest fruit yield of Jara lemon (1.35 t ha⁻¹). In consistent with the present results, Segarra-Carmona et al. (2010) found that the highest concentrations of spinosad and Bacillus thuringiensis caused 100% mortality in the third instar larvae of lemon butterfly. Iordanou and Charalambous (1998) evaluated some insecticides including spinosad for the management of citrus leaf miner in the laboratory. They found that spinosad gave satisfactory protection to the infestation of leaf miner for more than one-month period. The present results are consistent with the previous results of Planes et al. (2015) who found that spinosad and chlorfenapyr markedly reduced the nymphal population of citrus thrips, Pezothrips kellyanus and showed a significant reduction in fruit infestation. Besides above, Wang et al. (2009) found in a laboratory study that the combination of lethal and sublethal effects of spinosad reduced the pest population dynamics of Helicoverpa armigera (Hübner) through notably decreasing its survival and reproduction, which is also consistent with the present results. From the above discussion, it may be concluded that spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid10 SG) are the most effective insecticides among all the evaluated approaches with respect to highest leaf infestation suppression and highest fruit yield of Jara lemontree. The two treatments spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid10 SG) gave the highest MBCR (11.50: 1.0). Although it was found that spinosad (Tracer 45 SC) and chlorfenapyr (Intrepid10 SG) are highly effective biopesticides against lemon butterfly larvae, spinosad could be more preferable to chlorfenapyr as it is a less toxic, biologically derived material which is produced by fermented culture of the actinomycete Saccharopolyspora spinosa, a bacterial organism isolated from soil. Thus it would be more safer for environment and human health. Biopesticides can also provide ecologically sound solutions to pest problems and are known to be effective in suppressing pests that have already developed resistance to chemical pesticides. These biopesticides leave little or no

toxic residues and are commonly harmless to beneficial insects and other non-target organisms. It can be suggested that in order to suppress the infestation of lemon butterfly larvae, the citrus grower may spray spinosad for the effective management of larvae. Based upon cost-effectiveness and environmental safety issues, spraying of spinosad (Tracer 45 SC) @ 1.25 ml L⁻¹ of water would be the most effective biopesticide for the management of lemon butterfly larvae infesting Jara lemon trees in nurseries and gardens in Sylhet and other regions of Bangladesh where citrus plants are cultivated.

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